UltraScan User's Guide Standard Series

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Preface

About this Guide

This *UltraScan User's Guide Standard Series* is written to provide a brief description of the camera's operating principles, instructions on how to install the camera, and an introduction to imaging in DigitalMicrograph. For additional discussion of image processing, refer to the DigitalMicrograph User's Guide.

Preview of this Guide

The *UltraScan User's Guide Standard Series* includes the following chapters*:*

Chapter 1, "Introduction," summarizes the features of the *UltraScan* family cameras.

Chapter 2, "Getting Started," shows the first steps necessary to start the DigitalMicrograph software and prepare for the acquisition of an image (Gain Normalization and Magnification Calibration).

Chapter 3, "Image Acquisition," shows the user the most often used floating windows in DigitalMicrograph and a brief description on how to use them to view, acquire and save an image.

Chapter 4, "Quick Reference," is a brief summary of the steps necessary to operate a Gatan CCD camera.

Chapter 5, "Care and Maintenance," contains some general tips on the care and maintenance of the UltraScan camera. It also contains a list of questions that are often asked by users.

Appendix

- Hardware description provides the system requirements and a checklist. A brief description of the basic working principles of a CCD camera is also given. The pictures of different parts of the camera (camera head, back and front of the controller, etc.) are shown.

- CE Requirement.

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Table of Contents

ULTRASCAN USER'S GUIDE STANDARD SERIES **1**

PREFACE ..... **I**

ABOUT THIS GUIDE I
PREVIEW OF THIS GUIDE I
DISCLAIMER II
COPYRIGHT AND TRADEMARKS II

SUPPORT ..... **III**

CONTACTING GATAN TECHNICAL SUPPORT III

1 INTRODUCTION **1-1**

2 GETTING STARTED **2-1**

2.1 THE WORKPLACE 2-1
2.2 CAMERA CONFIGURATION 2-2
2.3 GAIN NORMALIZATION 2-4
2.4 MAGNIFICATION CALIBRATION 2-5
2.4.1 Low magnification *2-7*
2.4.2 "High" magnification *2-9*

3 IMAGE ACQUISITION **3-1**

3.1 VIEW 3-1
3.2 IMAGE NOTES 3-3
3.3 TEM SESSION INFORMATION (GLOBAL INFO) 3-4
3.4 IMAGE ACQUISITION 3-4
3.5 IMAGE SAVING AND PRINTING 3-5
3.5.1 Choosing directory *3-5*
3.5.2 Construct Image File Name *3-6*
3.5.3 Report Options *3-7*
3.5.4 Report *3-9*
3.5.5 Printing dimensions *3-10*

4 QUICK REFERENCE **4-1**

5 CARE AND MAINTENANCE **5-1**

5.1 GENERAL PRECAUTIONS 5-1
5.1.1 Peltier cooler maintenance *5-1*

5.1.2 Scintillator maintenance and precautions *5-2*
5.2 MOST OFTEN ASKED QUESTIONS 5-4

APPENDIX 1 I

HARDWARE DESCRIPTION I

A1 SYSTEM REQUIREMENTS I
A2 DESCRIPTION II
A3 INSTALLATION V
A3.1 Pressure lines VI
A3.2 Mounting the camera VIII
A3.3 Water lines IX
A3.4 Electrical connections XII
A3.5 Shutter-control output XIII
A3.6 Shutter-override output XIII

APPENDIX 2 XIV

CE CERTIFICATION XIV

DECLARATION OF CONFORMITY XV

INDEX XVI

List of Figures

FIGURE 2-1 DIGITALMICROGRAPH WORKPLACE 2-2
FIGURE 2-2 (A) CAMERA MENU (B) CAMERA CONFIGURATION 2-3
FIGURE 2-3 CAMERA MENU 2-5
FIGURE 2-4 GLOBAL MICROSCOPE INFO WINDOW 2-6
FIGURE 2-5 MICROSCOPE MENU 2-6
FIGURE 2-6 CROSS GRATING SAMPLE 2-7
FIGURE 2-7 INSTRUCTIONS WINDOW 2-7
FIGURE 2-8 CALIBRATION WINDOW 2-8
FIGURE 2-9 CHOOSE YES TO COMPLETE THE CALIBRATION 2-8
FIGURE 2-10 MAGNIFICATION TABLE 2-9
FIGURE 2-11 HIGH-RESOLUTION IMAGE OF A CRYSTALLINE SAMPLE 2-9
FIGURE 2-12 DIFFRACTOGRAM 2-10
FIGURE 2-13 HIGH MAGNIFICATION CALIBRATION INSTRUCTIONS 2-10
FIGURE 2-14 CALIBRATION WINDOW 2-11
FIGURE 3-1 IMAGE WINDOW 3-2
FIGURE 3-2 FOCUS LOUPE 3-3
FIGURE 3-3 IMAGE INFO 3-4
FIGURE 3-4 CAMERA ACQUIRE WINDOW 3-5
FIGURE 3-5 SAVING WINDOW 3-5
FIGURE 3-6 SAVING SET-UP WINDOW 3-7
FIGURE 3-7 SAVING DATA BAR 3-7
FIGURE 3-8 CUSTOM ITEMS 3-9
FIGURE 3-9 (A) REPORT VIEW (B) IMAGE VIEW 3-10
FIGURE 3-10 CONTROL PALETTE 3-11
FIGURE A1 `ULTRASCAN™ *STANDARD* SERIES CAMERA MOUNTED ON A TEM COLUMN. III
FIGURE A2 CONTROLLER'S FRONT PANEL III
FIGURE A3 CONTROLLER'S BACK PANEL IV
FIGURE A4 US 4000 SP BLOCK DIAGRAM. V
FIGURE A5 CAMERA INTERFACE DETAILS. VII
FIGURE A6 WATER LINES XI
FIGURE A7 ELECTRICAL DIAGRAM. XII

1 Introduction

The Gatan UltraScan™ family of cameras is designed to deliver the ultimate quality in transmission electron microscope images. It is the first digital camera system with the potential to replace film as a recording medium in the TEM. New technology has been introduced at every stage of the imaging process to improve resolution, sensitivity, dynamic range, linearity and speed of operation. The UltraScan™ family shares a compact mechanical design for bottom mount CCD (charged-coupled device) cameras with full retraction capability and complete compatibility with GIF and ENFINA™ spectrometers.

The UltraScan™ family of cameras is available in two series, *Reference* and *Standard* series. The common feature to these two series is the utilization of the patented High-Contrast Resolution (HCR™) optical technology. HCR™ technology results in images with higher resolution and higher contrast than images acquired with traditional optics and takes digital imaging resolution to a level that challenges film. The *Reference* Series UltraScan™ cameras incorporate the First Light™ readout electronics which reads out all four CCD output channels in parallel for increased speed while maintaining low-noise performance. The *Standard* Series UltraScan™ cameras read the CCD through a single ultra-clean low-noise channel. The *Reference* series features 2k x 2k and 4k x 4k CCD format and the *Standard* series features only the 4k x 4k format.

This user guide is written for the *Standard* series.

The intent of this guide is to:

- Make the user become proficient with the Gatan DigitalMicrograph software to correctly acquire images from the CCD camera.
- Give the user a brief understanding of the working principles of the camera.
- Allow easy troubleshooting.

2 Getting Started

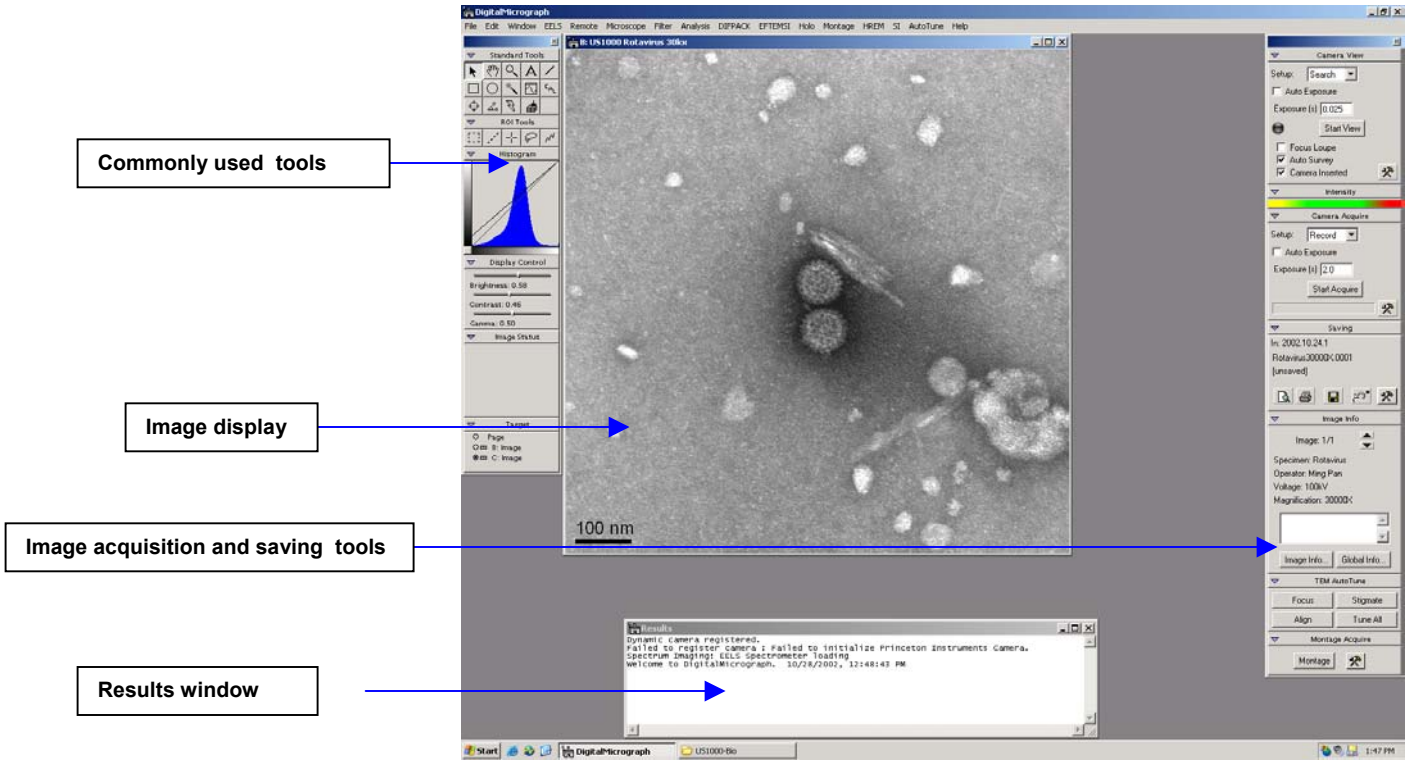
The following steps will enable the first-time user to switch on the digital camera, launch DigitalMicrograph and prepare for the acquisition of an image.

- Make sure that the viewing chamber is at the proper vacuum.
- Make sure the water and the three color-coded pressure lines are correctly connected to the camera and proper water pressure is maintained.
- Switch on the CCD camera controller.
- Flip the Peltier Cooler switch to the COOL position. For best results it is recommended waiting (2 hours or more) for the system to stabilize.
- Turn the computer on (if it is not on yet) and double click in the DigitalMicrograph icon to launch the application.

2.1 The Workplace

When DigitalMicrograph is launched, you will see the commonly used tools (left side), the Image Acquisition and Saving tools (right side), and the Result window as shown in Figure 2-1. These tools are composed by "floating" windows fully stackable to allow customization.

Figure 2-1 **DigitalMicrograph Workplace**



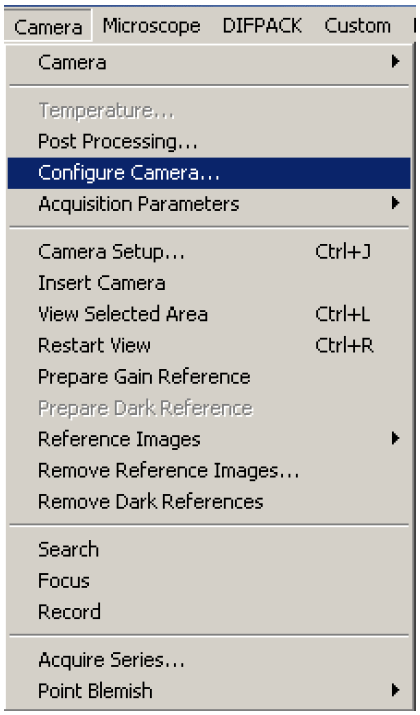
2.2 Camera configuration

If the camera is already configured by a Gatan service engineer, this section can be ignored.

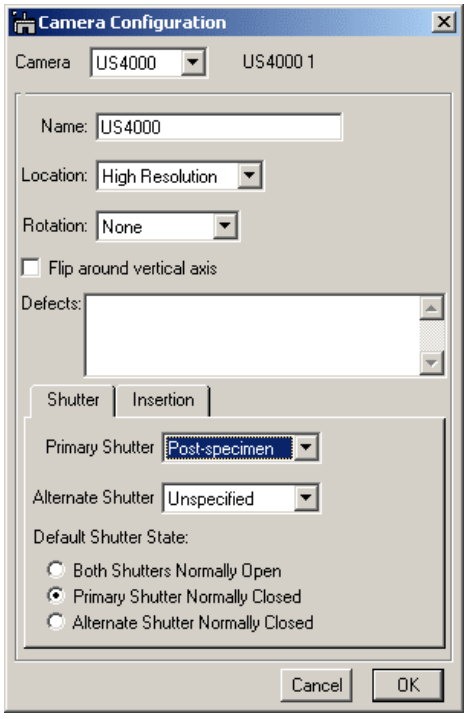
Before starting acquiring images, it is necessary to configure the camera. This process is shown in Figure 2-2.

Figure 2-2

(a) Camera Menu



(b) Camera configuration



- Select **Configure Camera** in the **Camera** menu (Figure (2-2a).
- In the Camera configuration dialog box (Figure 2-2b), you can change the name of the camera by typing in the **Name** field.
- Image rotation can be specified in the **Rotation** pull-down menu.
- The coordinates for column defect correction are entered in the **Defects** field. For example, entering the following:

  r0-8, r4088-4095, c0-8,c4088-4095

  (r and c indicates row and columns respectively) corrects for the 8 pixels wide edge which does not contain active CCD pixels.
- It is important to specify the **Primary** and **Alternate** (if there is one set up) shutter available to the camera.
- Finally you should specify the shutter status, i.e. the state of the shutter when the CCD camera is not acquiring any images. **Shutter Normally Closed** is the default setting.

2.3 Gain Normalization

It is important to prepare a new gain reference image after the CCD has reached the equilibrium temperature (normally this takes about 1-2 hours). The gain reference image should be checked regularly.

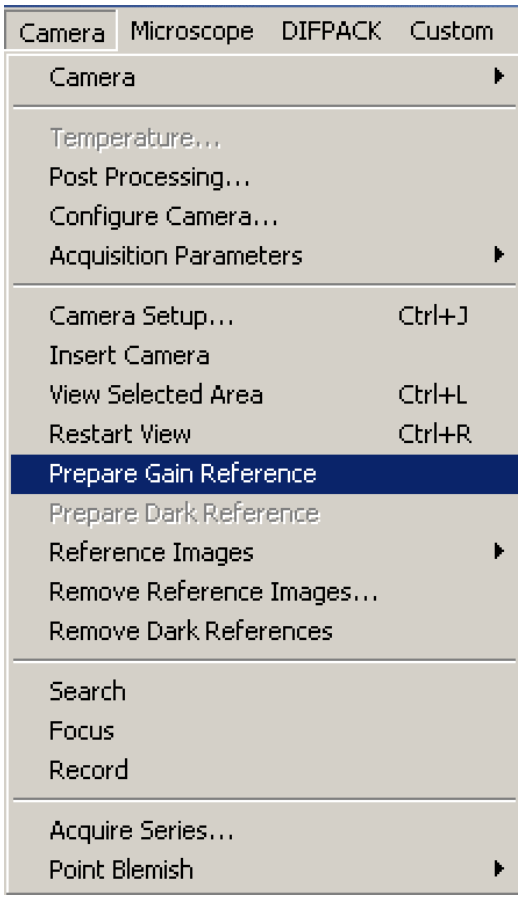
To prepare gain reference image, remove specimen from the field of view, evenly spread the illumination across the CCD sensor.

Under the **Camera** menu (Figure 2-3), choose **Prepare Gain Reference**. Set the target intensity to the value of 3000-4000. Normally it is sufficient to set the frames to average to 4. Then simply follow the instructions on the screen.

It is recommended to perform this operation when

- The camera temperature is stable (after at least two hours of cooling),
- Every time the camera is switched on,
- Once a week if the camera is always on, or
- Before acquiring any important images.

Figure 2-3 **Camera menu**



2.4 Magnification Calibration

The displayed nominal magnification on TEM is for photographic film and has an accuracy of 5-10%. The CCD camera is located on a different plane (height wise) respect to the film camera. As a consequence, the magnification must be calibrated. The calibration is done using standard calibration samples.

At “Low” magnifications:

- Use “cross grating sample” or any sample having known spacing

At “High” magnifications:

- Use “graphite” or any crystalline samples having known lattice spacing
- Use the FFT method.

It is very important to make sure DigitalMicrograph software correctly reads the TEM magnification. If the communication between the computer and the TEM is established, the magnification is read automatically. Otherwise, please

make sure DigitalMicrograph is set to prompt the user to enter a value for TEM magnification every time an image is to be acquired. This can be set by choosing the "Global Microscope Info" window shown in Figure 2-4 under the Microscope menu (Figure 2-5).

Figure 2-4 **Global Microscope Info window**

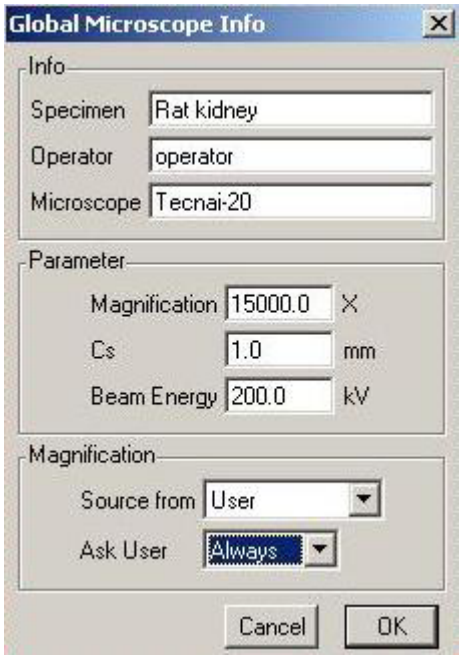
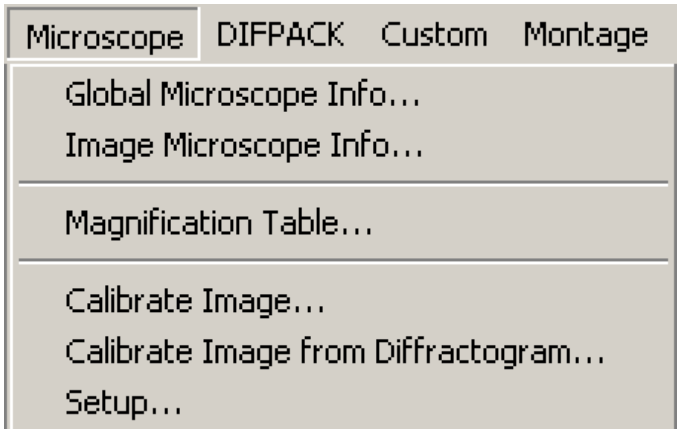


Figure 2-5 **Microscope menu**



In the "Magnification" section in the "Global Microscope Info" window (Figure 2-4) choose:

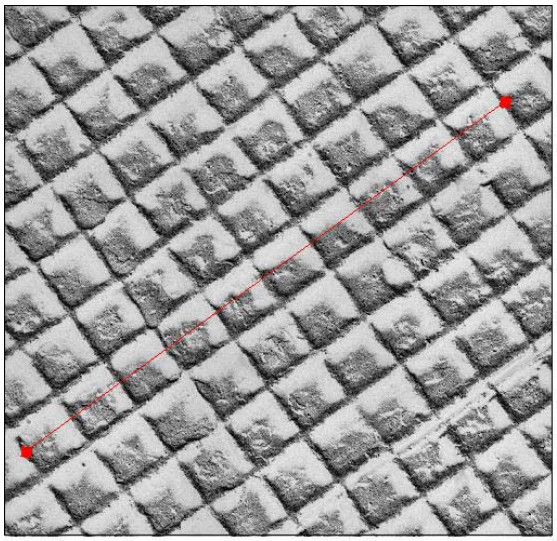
- Source from: **User**

- Ask User: **Always**
- Click **OK**

2.4.1 Low magnification

- Record an image of a cross grating sample (Figure 2-6)

Figure 2-6 **Cross grating sample**



- Choose “Calibrate image…” under the “Microscope” menu (Figure 2-5).

Follow the on screen instructions (Figure 2-7)

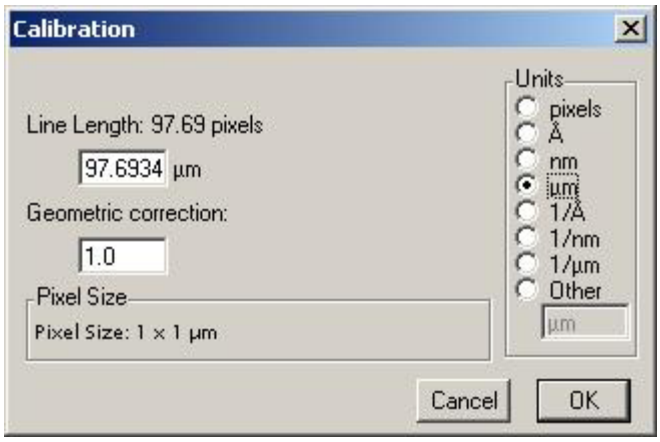
Figure 2-7 **Instructions window**

A red line will appear on the image.

- Position it on a feature of known size.
- Press OK on the “Calibrate image” window.

- Enter the correct distance for the selected feature (for example 10 line pairs of cross grating sample where the distance = 10 x 0.463µm) in the "Calibration" window and select the units (see Figure 2-8).

Figure 2-8 **Calibration window**



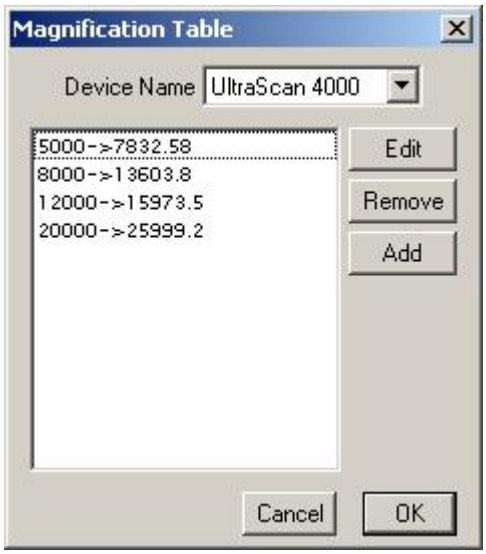
- Press OK.
- Choose YES in the next window (see Figure 2-8) to complete the magnification calibration.

Figure 2-9 **Choose YES to complete the calibration**

The calibration can be checked on the calibration table containing pairs of value: the nominal microscope magnification and the calibrated value. To view the **magnification table**:

- Select "Magnification table" (Figure 2-10) from the "Microscope" menu (Figure 2-5).

Figure 2-10 **Magnification table**

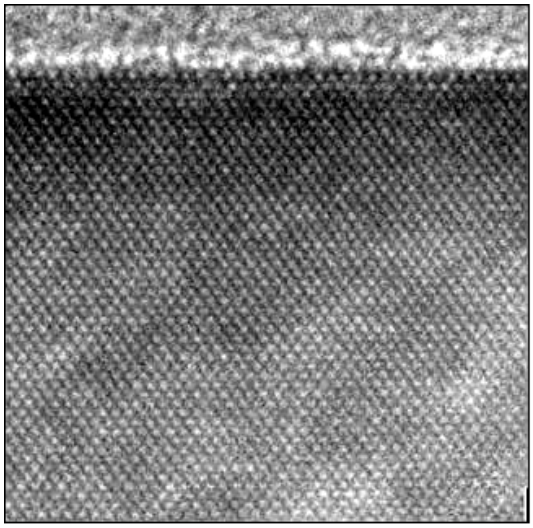


The first column represents the nominal TEM magnification and the second column is the actual magnification at the CCD.

2.4.2 "High" magnification

- Record a lattice image of the crystalline sample (Figure 2-11).

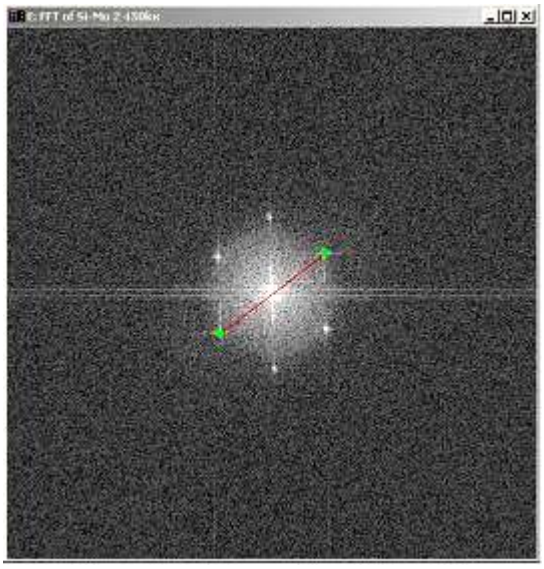
Figure 2-11 **High-resolution image of a crystalline sample**



- Choose "Calibrate image from Diffractogram…" under the "Microscope" menu (Figure 2-5).

A diffractogram will be calculated (Figure 4-12).

Figure 2-12 **Diffractogram**



- Follow the on screen instructions (Figure 2-13)

A red line will appear on the diffractogram.

- Position the endpoints of the red line on two symmetrical diffraction peaks (Figure 2-12).
- Press OK to specify the reciprocal unit and the d-spacing (in the corresponding real units) in the next window (Figure 2-14).

Figure 2-13 **High magnification calibration instructions**

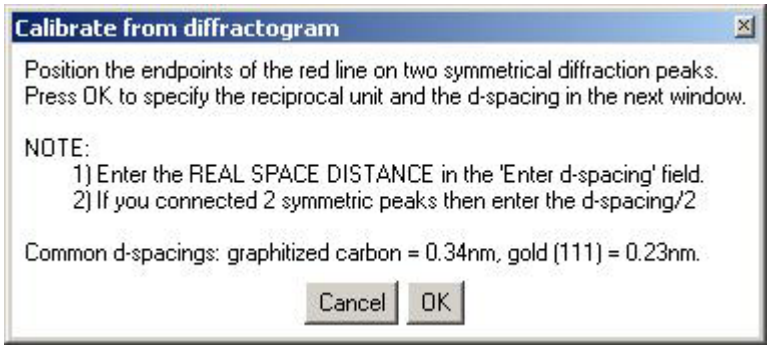
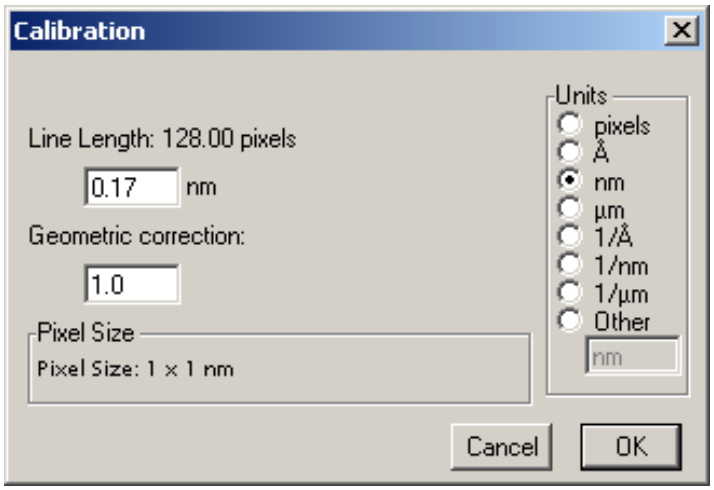

Calibrate from diffractogram
Position the endpoints of the red line on two symmetrical diffraction peaks.
Press OK to specify the reciprocal unit and the d-spacing in the next window.
NOTE:
1) Enter the REAL SPACE DISTANCE in the 'Enter d-spacing' field.
2) If you connected 2 symmetric peaks then enter the d-spacing/2
Common d-spacings: graphitized carbon = 0.34nm, gold (111) = 0.23nm.
Cancel
OK

Figure 2-14 **Calibration window**



3 Image Acquisition

This chapter contains some instructions on how to acquire an image and a description of the most frequently used floating windows.

Now you are ready to:

- Insert the sample,
- Find a region of interest
- Start using the CCD camera to view your specimen.

3.1 View

The view mode (shown in Figure 3-1) allows user to observe **Live** (continuous) images on the monitor. The setup has two operation modes which are preset for easy use:

- The **Search** mode displays a continuous (low resolution) image with the entire CCD field of view. This is to allow the user to quickly conduct a sample search using the CCD camera.
- The **Focus** mode displays a continuous image at higher resolution than the **Search** image. Fine adjustment of focus setting can be done with this operation mode.

In addition, there is another way to adjust the image focus. Click on the **Focus Loupe**. This allows the user to focus the image by using a small image area with high speed. The size of the box can be resized by dragging its corner and its location can be easily changed by putting the pointer inside the box and dragging it. The **Auto Survey** updates the image display in real time.

Auto Exposure is enabled and disabled by clicking on the **Auto Exposure** selection box. Since a test exposure must be made regularly to ensure proper exposure time for the beam intensity on the CCD and for 4kx4k CCD, this means 16million pixels must be read out each time, it is recommended to turn off this feature. When the box is not selected the user is in control of the exposure time that can be changed by either typing in the **exposure time** box and then pressing the Enter key, or by hitting the up and down arrow keys to respectively double or halves the exposure time.

To view a continuous image of the sample:

- Select the **Camera inserted** box or make sure it is selected.
- Setup: **Search** mode
- Press **Start View** to start the image acquisition

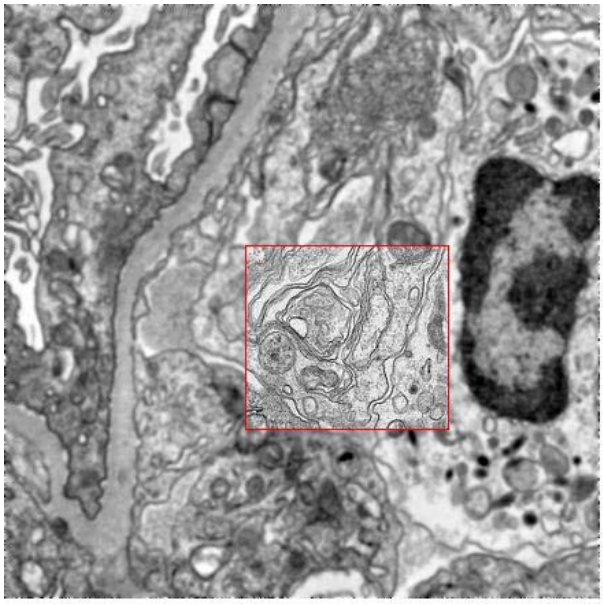
If you need to better adjust the microscope focus on the sample:

- Click **Focus Loupe** in the Camera View window.

Only the selected area (see Figure 3-2) will be imaged at a fast rate allowing to set a more precise focus on the area of interest.

Figure 3-1 **Image window**

Figure 3-2 Focus Loupe



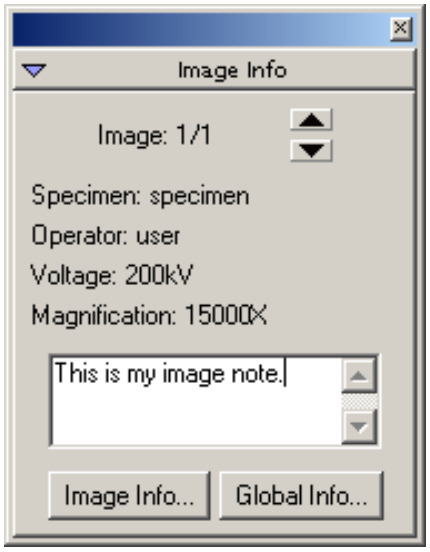
The acquisition can be stopped by:

- Pressing **Stop View**, or
- Pressing the Spacebar.

3.2 Image Notes

The user can type in notes in the **Image Info** window shown in Figure 3-3. Saving an image will automatically save the image notes with the image file. The **Up/down arrows** allow user to browse through the images saved in a session when using the AutoSave function (Section 3.5 below).

Figure 3-3 Image Info



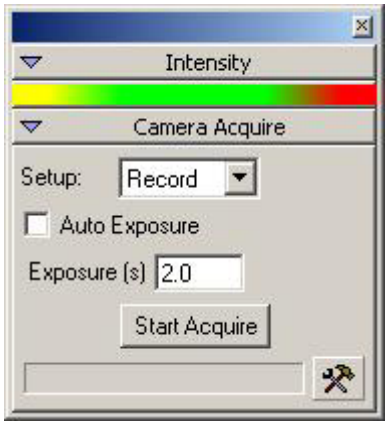
3.3 TEM Session Information (Global Info)

Before acquiring any images, it is recommended to enter the correct information about the sample, operator, TEM name, etc. Click on the **Global Info** button (Figure 3-3) to type in all these information. This function can also be accessed under the Microscope menu. The global information is saved automatically with every image that is acquired.

3.4 Image Acquisition

To record the final image, click on the **Start Acquire** button in the Camera Acquire palette shown in Figure 3-4. This mode is preset to acquire a full resolution CCD image. As explained previously, it is recommended to turn off the **Auto Exposure** feature for 4kx4k CCD.

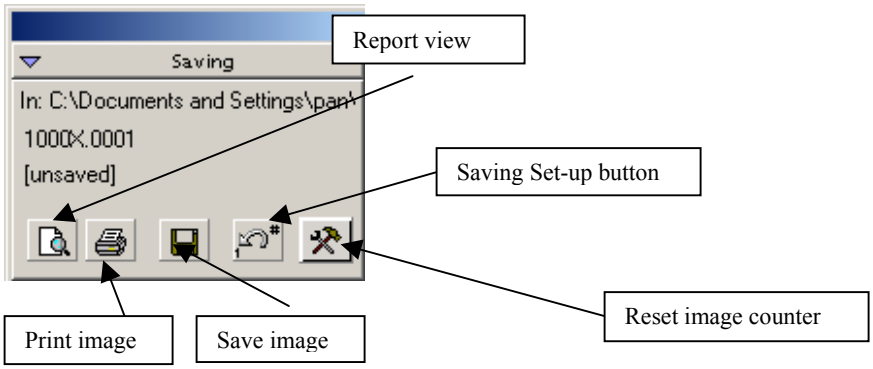
Figure 3-4 Camera Acquire window



3.5 Image Saving and Printing

The recorded images are displayed on the monitor. Pressing on the floppy disk icon or on the printer icon in the **Saving** palette (Figure 3-5) the image can be saved or printed respectively. Images can be easily saved in any user specified directory with a common root name.

Figure 3-5 Saving window



3.5.1 Choosing directory

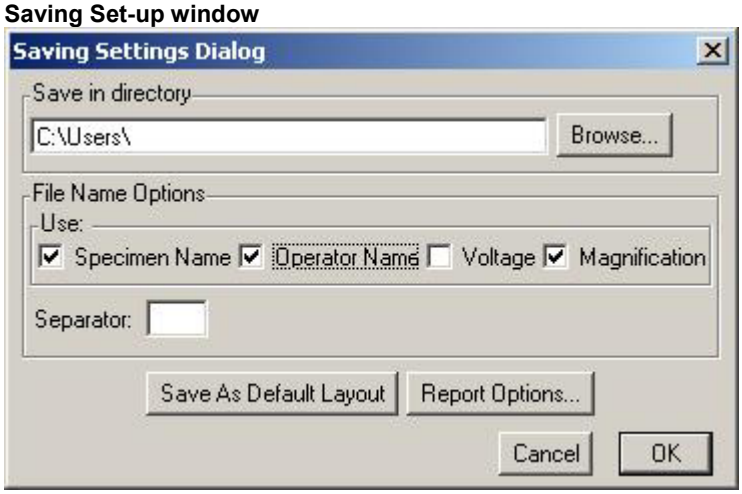
Click on the **Set up** button in the **Saving** window (Figure 3-5). The **Saving Set up** dialog box will be displayed as shown in Figure 3-6.

In the **Save in directory** (Figure 3-6)**,** click on the **Browse** button. Select or create a directory for saving images.

3.5.2 Construct Image File Name

In the **File Name Options** field**,** specify the way you want the image file name to be constructed.

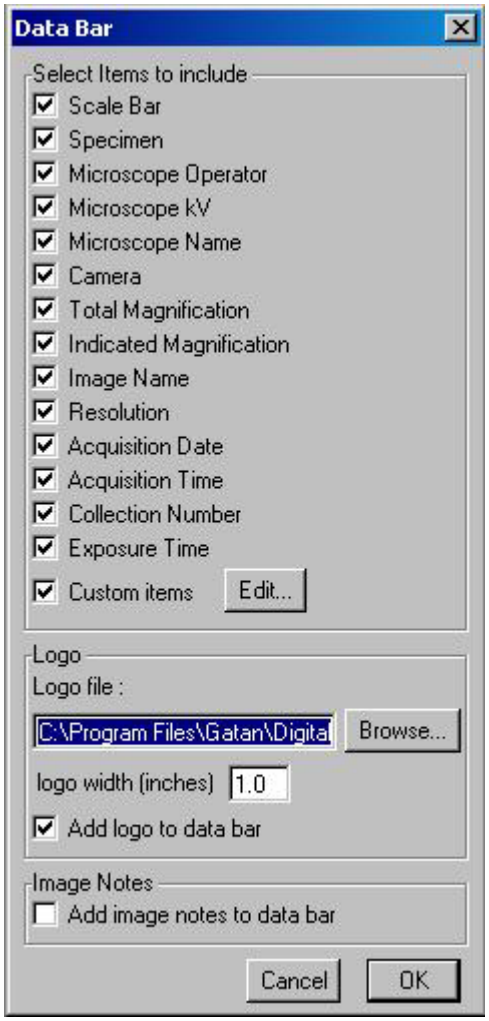
Figure 3-6 **Saving Set-up window**



Report Options

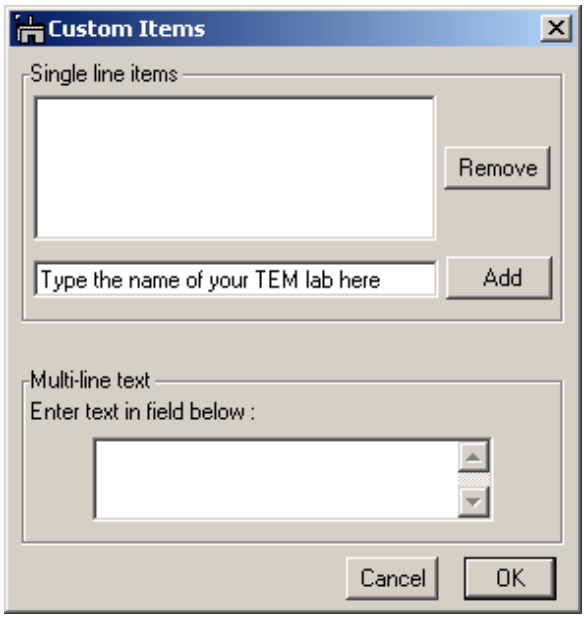
Click on the Report Options **button to view the dialog box shown in Figure 3-7. You can choose to have any parameters listed to be included in your report. You can also add a logo file and image notes to the report.**

Figure 3-7 Saving Data Bar



Custom item allows user to add text information to every report page. This feature is useful to add the name of your institution, or the name of your TEM lab. Click **Edit** to get the **Custom items** dialog box as shown below (Figure 3-8).

Figure 3-8 Custom Items



Type the text in the **Single line item** (for example the name of your TEM lab) and then click the **Add** button. Now the text is entered in the window of the **Single line item**. Click **OK**.

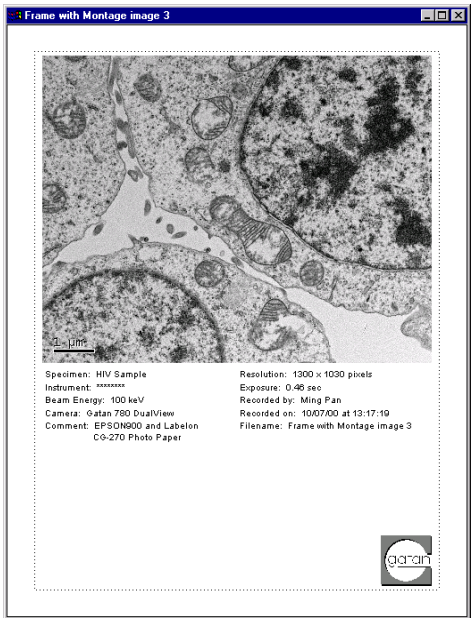
The style of the **Custom item** can be easily changed in the **Report page** (next section).

3.5.3 Report

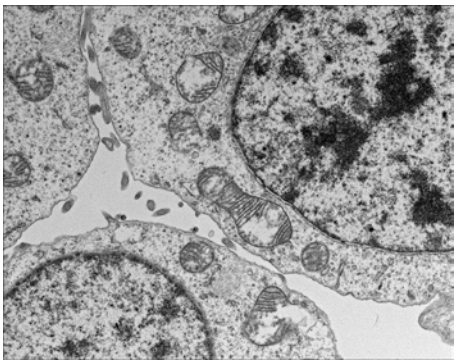
The report button (the one with a page and a magnifier on the Save palette in Figure 5-5) will toggle between report view (Figure 5-8 a) and image view (Figure 5-8 b). In the report view, an image is displayed of the print preview mode with parameters as specified in the **Report Options**.

Figure 3-9

(a) Report view


Frame with Montage image 3
Specimen: HIV Sample
Instrument: ********
Beam Energy: 100 keV
Camera: Gatan 780 DualView
Comment: EPSON900 and Labelon CG-270 Photo Paper
Resolution: 1300 x 1030 pixels
Exposure: 0.46 sec
Recorded by: Ming Pan
Recorded on: 10/07/00 at 13:17:19
Filename: Frame with Montage image 3

(b) Image view



Users can customize the layout and selection of parameters on the page (for example, using pointer to move the text around) and then save the setting as a report template. This is done by clicking the **Save As Default Layout** button in the **Save setup** dialog**.**

The **Custom item** on the page can be changed to different text style to make it more visible. First position it on the **Report page** and then choose the font and size of your liking. Click

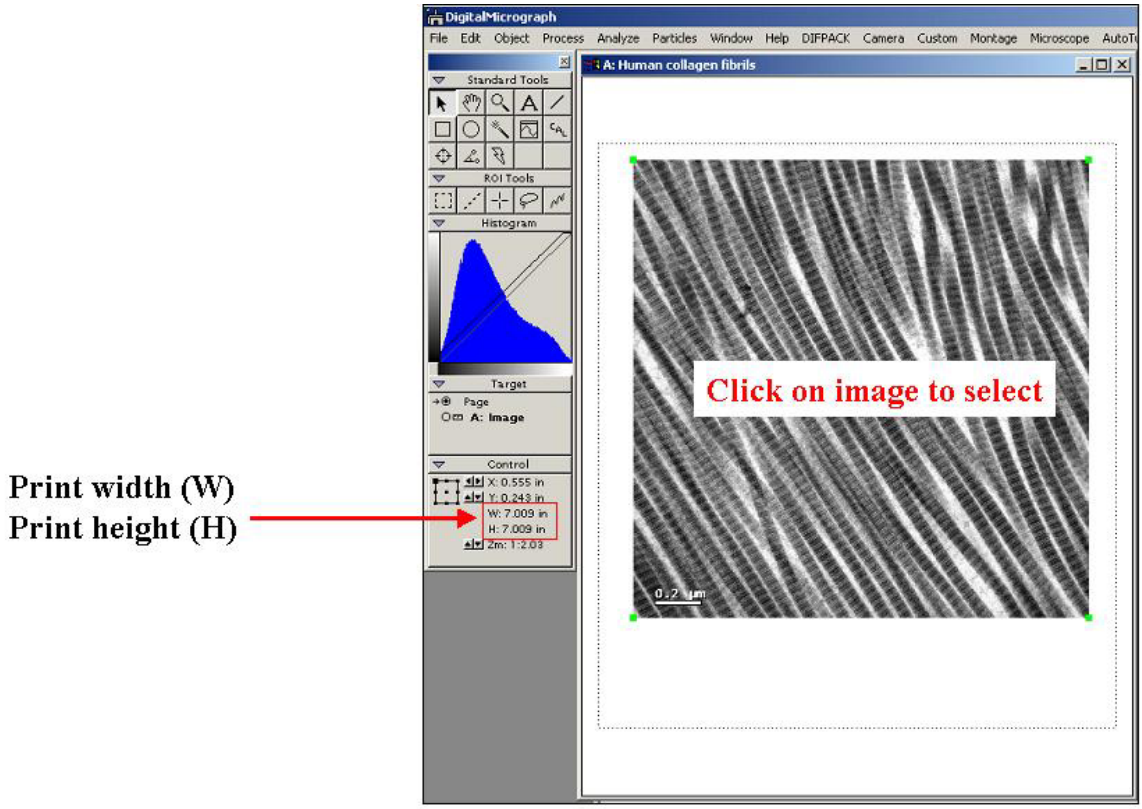
3.5.4 Printing dimensions

Digital images can not be magnified "indefinitely" since "Pixelation" may occur. To print non-pixelated images, ppi (pixel per inch) must be at least 144. For 1k x 1k CCD images, 144ppi means the images will be printed ~ 7 x 7 inch.

To find out the good printing dimensions for your image, use the **Control** palette in DigitalMicrograph (Figure 5-9).

- Select the image (click on it).
- Read the printing dimensions on the **Control** palette.
- Click on the printer icon in the **Saving** window (Figure 5-5) to print.

Figure 3-10 **Control palette**



4 Quick Reference

The steps necessary for operating Gatan CCD camera are summarized here.

- Make sure that the viewing chamber is at the proper vacuum.
- Make sure the water and the three color-coded pressure lines are correctly connected to the camera and proper water pressure is maintained.
- Switch on the CCD camera controller.
- Flip the Peltier Cooler switch to the COOL position. For best results it is recommended waiting (2 hours or more) for the system to stabilize.
- Turn the computer on (if it is not on yet) and double click in the DigitalMicrograph icon to launch the application.
- Check the TEM session information by clicking on the **Global Info** button on the **Image Info** palette (Figure 3-3).
- Prepare a gain reference image by choosing **Preparing Gain Reference** under the **Camera** menu (Figure 2-3).
- Click on "**Start View"** on the **View** palette in **Search** mode (Figure 3-1) to position sample area of interest. (disable **Auto Exposure).**
- Click on **Focus Loupe** on the **View** palette (Figure 3-1) to check the final focus. Make sure **Auto Survey** is checked.
- Click on **Start Acquire** button in the **Image Acquire** palette (Figure 3-4). (disable **Auto Exposure).**

- Click on **Report View** button on the **Saving** window (Figure 3-5) to view image in print preview mode.
- Click on **Save** button (diskette icon on the **Saving** window) (Figure 3-5) to save image in the designated folder.
- Click on **Print** (Figure 3-5) button (printer icon on the **Saving** window) to send image to printer.

5 Care and Maintenance

This chapter contains some general tips on the care and maintenance of your UltraScan camera. It also contains a list of questions that are often asked by users.

5.1 General Precautions

The following are some general precautions users should adhere to in the care and maintenance of their camera.

5.1.1 Peltier cooler maintenance

- The Peltier cooler should not be switched on if the cooling water is not flowing.

The Peltier cooler consumes about 12 W of power. If this heat is not removed by the cooling water, there is a possibility that damage to the Peltier cooler and/or the CCD may occur.

- Check the flow of cooling water periodically.

If the flow rate of the cooling water deviates significantly from the value originally set, make sure the lines are not obstructed and adjust the pressure regulator to bring the flow back to the original level. Remember, if the water flow stops while the Peltier cooler is on, damage to the camera head may result.

- The Peltier cooler should not be left on when the TEM cooling water is turned off or fails.

The Peltier cooler is cooled by water from the TEM chiller. If the Chiller is turned off or fails for a long period, there could be severe damage to the camera. Be aware that for some microscopes the cooling system turns off in the Stand By mode. Be sure the cooler of the camera is also turned off in that situation.

- Turn off the Peltier cooler if the microscope viewing chamber is to remain at air pressure for a long period (more than 30 minutes to an hour).

When isolated by the gate valve, the CCD vacuum is good for about half an hour, after which some frosting on the CCD may occur if the CCD is kept cold. A small amount of ice will sublimate away once high vacuum is restored in the viewing chamber and the CCD gate valve opens. Removing large amounts of condensation will require the electronics to be switched off for about 30 minutes after the vacuum in the viewing chamber is restored, so that the CCD warms up while it is in a good vacuum.

5.1.2 Scintillator maintenance and precautions

- Minimize exposing the scintillator to the electron beam when the camera is not in use.

It is particularly important to protect the camera from being unnecessarily exposed to an intense unscattered beam. As a general rule, if you avoid conditions which would saturate any part of the sensor area in less than 0.1 sec (about 10 nA/cm^2 at the sensor), the scintillator lifetime will probably exceed the lifetime of the rest of the system. On the other hand, each time any CCD pixels are intensely saturated, the scintillator efficiency may be permanently impaired for those pixels, resulting in a variation of the scintillator efficiency across the image field. If you are not using the camera, keep the viewing screen down or retract the camera to protect the scintillator from beam damage. Also, a retracted camera will not collect debris or abrasive particles that may fall down from the TEM's film-transport mechanism.

- Minimize exposing the scintillator/CCD to high beam intensity when acquiring diffraction patterns.

There is no defined cutoff where the scintillator will be damaged. Rather, damage can result from prolonged exposure of the scintillator to a very high beam intensity (above 4,000 counts in binned by 1 mode and 15,000 counts in binned by 2 and higher modes). If you have no saturation with a short exposure, then there is no danger. You can then increase the exposure time and still not harm the scintillator as it is the beam intensity on the scintillator that causes the problem and not the accumulated signal in the CCD.

It is recommended to use beam stop to block the central spot when acquiring SAED diffraction patterns.

If you think you have damaged the scintillator, take an unprocessed image of uniform illumination. If the spot turns out dark, you have damaged the scintillator! If it is bright, spread out the beam and set the illumination so you have about 10,000 counts/sec (in binned by 2 mode) and switch the shutter switch on the controller to Open. Expose the scintillator for 3 min, switch back to Auto, and take another picture. The bright area should be gone and the scintillator should operate normally.

- Warm the scintillator periodically.

Some microscopes have a significant vapor pressure of diffusion pump oil in the viewing chamber. Since the camera is maintained at temperatures below -20ºC during use, it can act as a cold trap and accumulate an oil film. While thin this film does not affect imaging, but if it becomes thicker or coalesces into droplets, stronger and stronger gain correction will become necessary. Other problems caused by the oil film are charging and the sticking of dirt which could otherwise be blown off easily. If left for too long, the hydrocarbons chains in the oil will be cracked by the electron beam making them non-volatile and un-dissolvable. Then it is necessary to send the camera back to have the scintillator serviced by Gatan. Fortunately, these problems can be avoided by adhering to the practice described below.

If you have a clean vacuum in the camera chamber area, warm the scintillator once or twice a month. But if you have backstreaming from your diffusion pump, you will need to do it once a week. The camera should only be warmed for 15 hours or overnight, but not longer. If the camera is used sufficiently without warming, the electron beam can harden the trapped oil making the scintillator difficult to clean.

Switch the Peltier cooler to the “Warm” position.

It is OK to run the Peltier cooler in the warm position overnight, but do not run it when the camera is at air pressure or if the cooling water is not flowing.

Check the vacuum system.

If oil accumulates even with regular warming of the scintillator, it is recommended that the vacuum system be checked. Note that the oil will not be imaged as soon at 120 kV as at a lower voltage (40 kV), so that a problem can exist for some time before it is noticed, by which time the beam might already have hardened the film. That fact can be used to diagnose the presence of oil if the user takes an image at a low kV periodically to check for the presence of oil.

Examine the surface of the scintillator.

If the surface of the scintillator becomes dirty or scratched, ship the camera to Gatan for service. Do not attempt to clean the scintillator yourself as damage can result.

5.2 Most Often Asked Questions

The following are some of the questions most frequently asked by camera users. If you are having a problem with your camera, check and see if the tips listed below are helpful.

1. Why does the US4000 SP camera saturate far below the 16,000 value (14-bit dynamic range) in 1x binning mode?

For US4000SP camera, the saturation of 2x binning readout is set to 14-bit, i.e. 16,000 value. The higher resolution 1x binning readout mode does not make full use of the full range of the output amplifier because of the smaller pixel size. The US4000SP saturates at roughly 1/3 of the full digitization range by intent.

2. There is no image on the CCD, what should I do?

Check if the cable from the UltraScan controller to the camera is properly connected.

Check the “Camera View” floating window to see if the “Camera Inserted” box is checked (camera inserted).

Check if the Shutter switch on the UltraScan controller is set to AUTO.

Check if the beam has drifted.

Check if the TEM viewing screen is lifted.

Check if you have a film plate stuck above the camera in the TEM.

You may have an image and not being able to see it. Move the pointer around to get an idea of the counts. If the value is over 4,000 counts (in 1x binning mode) the CCD is saturated. Reduce the beam intensity or shorten the exposure time.

Why do I have to blank the beam when the CCD is reading out?

You do not want to expose the CCD to a signal when it is reading out. It is like continuing to expose the film during its retraction. Blanking the beam will prevent any electrons from reaching the CCD.

3. How often do I have to acquire a gain reference?

Whenever the image quality seems to be deteriorating. The most reliable way to see if a new gain reference is advisable is to take a gain normalized image of uniform illumination with no specimen in the beam and look for any spurious pattern. If you see one, update the gain reference.

4. What is the shortest and/or the longest exposure time I can use?

The shortest time is actually determined by the shutter speed of the TEM shutter (about 0.025 sec). Exposures over 1 minute are unusual.

5. My images are noisy, what can I do?

Try increasing the exposure or current density of the beam.

6. My images are slowly deteriorating, what should I do?

Determine if the CCD is warming up. Check that the water lines are not clogged. Check the water temperature of the chiller- should be approximately 20-23 °C. Be sure to acquire a new gain-reference image.

7. The image is brighter (or darker) on the top than on the bottom, what should I do?

Check that the CCD has been cooled for at least 2 hours.

Check that the shutter switch on the controller is set to AUTO.

It could be a bad reference image. Retake another one.

Check that the shutter is functioning correctly. If it is, the shutter indicator light on the controller would go on and off as you take an image.

8. When viewing, the image sometimes gets suddenly bright/dark, what is happening?

Sudden changes in the contrast may be due to stray x-ray photons that saturate some of the pixels.

9. How long is the lifetime of the CCD?

None of Gatan's CCDs has failed due to old age so it is hard to answer. So far, there are no known mechanisms for aging.

10. How often and for how long should the camera be warmed?

If you have a clean vacuum in the camera chamber area, between once or twice a month is enough. But if you have backstreaming from your diffusion pump, you will need to do it more frequently. The camera should only be warmed for several hours or overnight, but not longer. A quick way to see if oil is accumulating is to take a uniform illuminated image at 40 or 60 kV.

11. The camera takes a long time to insert/retract, what should I do?

Check your compressed-air pressure. It should be between 30-60 N/cm^2 .

Then the pressure regulator on the red (retract) line should be adjusted so that insert and retract times are equal.

12. Why can't I expose photographic film with the camera on and inserted?

When the camera is on and inserted, the TEM shutter is controlled by the CCD camera software. In order to protect the CCD, the software (default

setting “shutter normally closed”) closes the TEM shutter if the CCD is not set to acquire any images. In this case, one cannot expose the photographic film.

To expose the film, one should (1) simply retract the CCD camera, or (2) force the shutter open with the switch on the front panel, or (3) set the shutter configuration to “shutter normally open”.

13. I see a ghost image that won’t go away, what should I do?

It is possible that the dark or gain reference has a ghost image. Try clearing and re-acquiring the reference images. You can remove the dark reference image by choosing *Remove dark references* under the Camera menu.

If the ghost image still exists, it is likely caused by exposing intense beam on the CCD. Spread out the beam and set the illumination so you have about 10,000 counts/sec (in 2 times binned mode) and switch the shutter switch on the controller to Open. Expose the scintillator for 2-3 min, switch back to Auto, and take another image.

Appendix 1

Hardware description

A1 System Requirements

To use the UltraScan *Standard* series CCD cameras, the following hardware and software systems are required:

Windows 2000 Professional or Windows XP Professional

Minimum 512 MB RAM (1GB RAM preferred)

40GB, 1GHz Hard-drive or better

Pentium III or later

Available microscope's bottom port

Water connections

Air connections

Gatan Microscopy Suite (GMS) software 1.1 or later

FireWire capability

A2 Description

An UltraScan™ *Standard* Series camera acquires an electron microscope image into a computer in four stages. First a scintillator converts the electron image into a high-resolution light image. Then a fused fiber-optic plate captures the light from each point in the image and channels it to the CCD. The CCD then converts the transferred light image into a second electron image. Finally, the CCD shifts that image charge row by row to the low-noise output amplifier, readout electronics and digitizer.

Sensitivity is achieved by maintaining high efficiency at each stage of the image signal chain: light generation in the scintillator, optical coupling by the fiber optic plate, image detection by the CCD pixel and also by cooling and low-noise readout. Resolution is achieved by minimizing light scatter in the scintillator and light leakage amongst fibers in the fiber optic plate and by optimized proximity focus at the end of the fiber plate through ultra-thin bonding technology. In addition, since the signal chain contains no moving or flexible parts the detection efficiency has very high stability over time. These techniques together constitute the HCR™ technology, which is at the heart of the UltraScan™ family.

Figure A1 shows the placement of an UltraScan™ *Standard* Series camera on a TEM column. The camera is mounted below the viewing and film chamber. The camera controller shown in Figure A2 (front panel view) and Figure A3 (back panel view) and an image-acquisition computer are linked to the camera by supplied cables. Compressed air for controlling the camera insertion and retraction is taken from the microscope as is the cooling water.

Figure A1 **`UltraScan™ *Standard* Series camera mounted on a TEM column.**

Figure A2 **Controller's front panel**

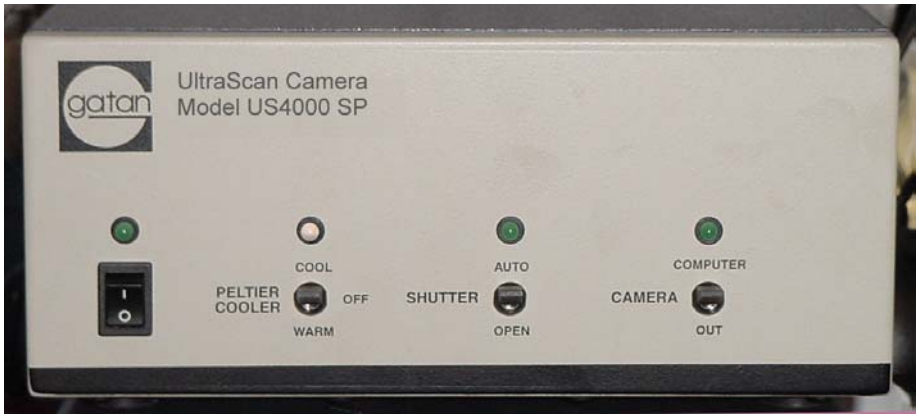


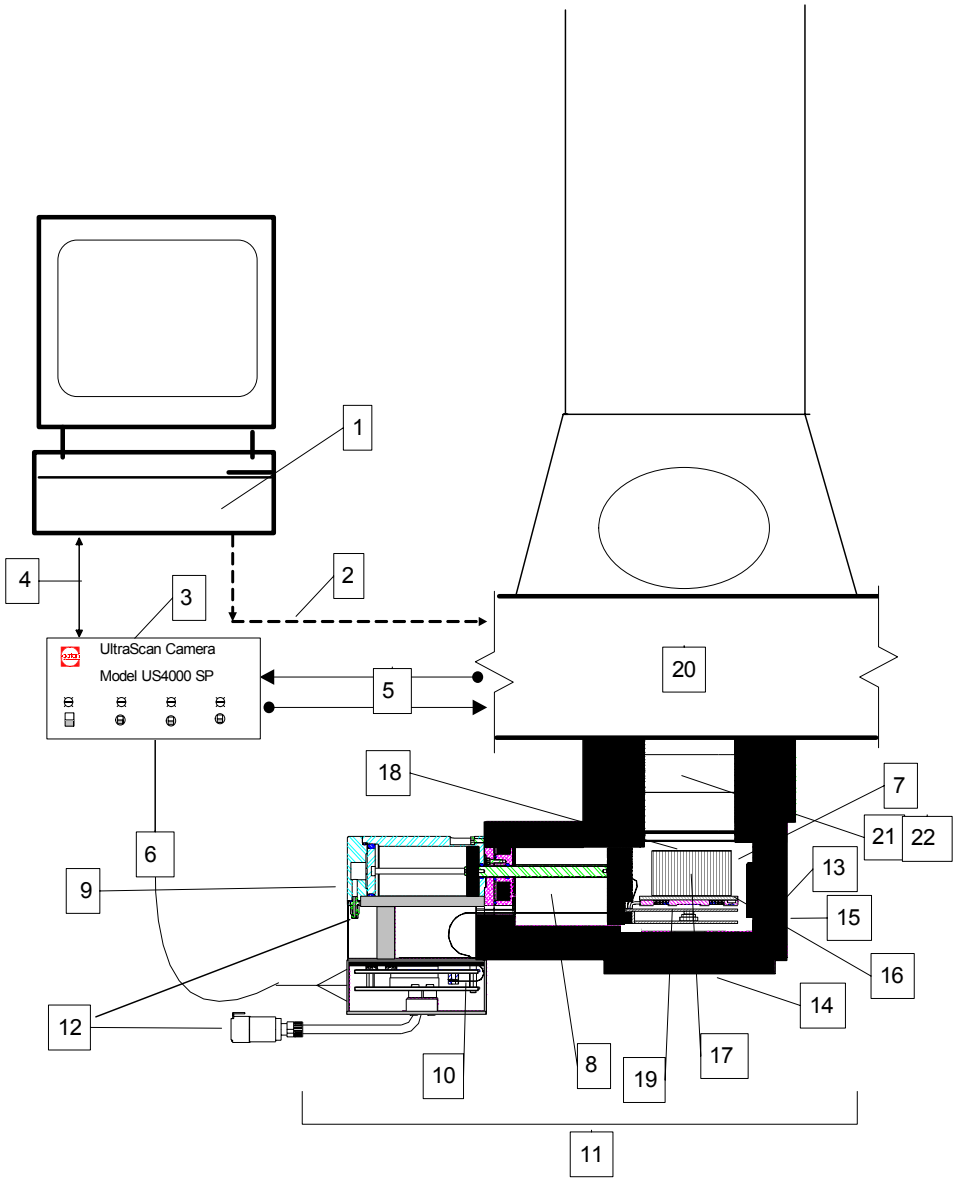
Figure A3 **Controller's back panel**

Figure A4 is a schematic illustrating the UltraScan *Standard* Series system. The system is composed of a retractable CCD camera coupled to an electron scintillator through a fiber optic plate, a controller/digitizer unit, a computer for image storage and processing, cables, and application software. The camera can be cooled down to -20 °C and can be inserted into or retracted from the electron beam by a software command or by a switch onto the controller's front panel (Figure A2).

Figure A4 **US 4000 SP block diagram.**

UltraScan System

1. Computer
2. TEM-interface cable (optional)
3. Controller
4. Camera-interface cable
5. Shutter-interface cable
6. Camera-interface cable
7. UltraScan vacuum chamber
8. Drive assembly
9. Pneumatic inlets
10. Preamp assembly
11. UltraScan camera module
12. Coolant connections
13. X-ray shielding
14. Blanking flange
15. Accessory port
16. Charge-coupled device
17. Fiber optic plate
18. Electron scintillator
19. Peltier cooler
20. Electron microscope
21. Microscope vacuum
22. UltraScan flange



A3 Installation

The following steps will enable the first-time users to install the camera with ease and safety:

- Unpack the camera, the camera electronics, and the computer.
- Place the controller within 10ft. (3m) of the column.
- Place the computer anywhere that is convenient.

- Place the gray camera cable between the controller and the knee well of the microscope.

Place with the female end towards the camera.

A3.1 Pressure lines

The camera needs to be connected to the microscope's pneumatic system (Figure 2.5). This will allow controlling the insertion and retraction of the camera. The camera head can be:

- Positioned **fully in** with the scintillator exposed to the electron beam and the CCD inside the microscope vacuum.

- **Partially retracted** with the scintillator out of the beam but the CCD still under the microscope vacuum.

- **Fully retracted** with both the scintillator and the CCD sealed inside a separate vacuum enclosure.

The air kit, supplied with the camera, consists of three sets of tubes (two per set), numbered 1 through 6 in Figure 2-5. Tubes 1 and 4 are green, 2 and 5 are yellow, and 3 and 6 are red. Tubes 1 through 3 are thinner than tubes 4 through 6. All tubes have male quick-disconnect fittings except for 5 and 6, which have female quick-disconnect fittings. Tube 6 has a pressure regulator along its length. Three Ts are used to attach the tubes to the microscope's pneumatic system.

- Attach tubes 1 through 3 to the pneumatic inlets on the camera head. (Going clockwise, the order of the tubes is 1,2, and 3).

- Cut the compressed-air line going to the diffusion pump gate valve of the microscope pneumatic system.

  The cut must be made as cleanly as possible. It can be made anywhere along the compressed air line.

- Connect the two parts of the compressed-air line together in series with a T.

  The two inline fittings of the T must be of a compatible size to the air line. As the lines of different microscopes may have different diameters, it is a good idea for the installer to have his own Ts at hand in case the supplied Ts do not fit. The perpendicular fittings of the supplied Ts fit tubes 4 through 6. If the installer's Ts are used, their perpendicular fittings must also fit tubes 4 through 6.

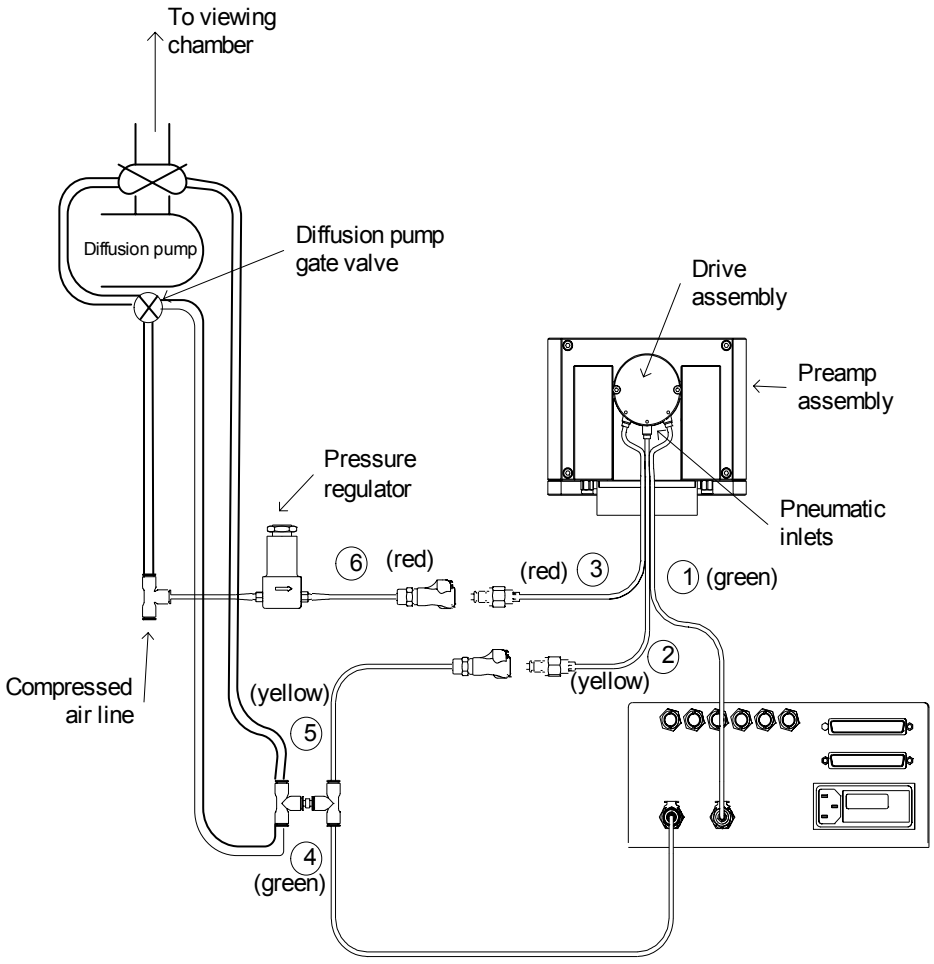
- Connect tube 6 to the perpendicular fitting of the T along the compressed-air line.

- Cut the open line to the diffusion pump gate valve of the microscope's pneumatic system in two places.

The cuts must be made as cleanly as possible. They can be made anywhere along the Open air line, but it is ideal for the first cut to be made one third of the line's length from the diffusion pump gate valve and the other cut two thirds of the line's length away from the valve.

- Connect the three sections of the Open line together in series with two Ts.

Figure A5 **Camera Interface details.**



- Connect tubes 4 and 5 to the remaining two fittings on the Ts along the open line.
- Connect tubes 2 and 3 to tube 5 and 6 respectively through their quick-disconnect fittings.
- Connect the male quick-disconnect fitting of tube 1 to the outlet on the backside of the controller labeled **Air Out** (Figure 2-3).
- Snap the male quick-disconnect fitting of tube 4 into the inlet on the backside of the controller labeled **Air In** (Figure 2-3).

The quick disconnect fittings automatically seal the lines off when not connected to anything, so that they may be connected and disconnected from the controller or the microscope without affecting the operation of the microscope.

A3.2 Mounting the camera

If you are mounting the camera to the microscope:

- Vent the microscope's viewing chamber following the microscope manufacturer's instructions.
- Remove the blanking flange on the microscope's on-axis camera port beneath the film chamber. Follow the instructions given by the microscope manufacturer.
- Inspect the o-ring seal to the microscope for dust and fibers.

  The seal must be clean in order to maintain the vacuum. If necessary, clean and lightly coat the o-ring with vacuum grease.

- Mount the camera to the microscope.

  Make sure that the sealing surfaces are clean.

- Reverse the end-point connections for tubes 1 and 3 from the normal configuration shown in figure 2-5.

  Plug tube 3 into the **Air Out** outlet on the backside of the controller (Figure 2-3) and plug tube 1 into tube 6 (pressure-regulator line). This is a reversal of the normal configuration but will ensure that when the diffusion pump gate valve is closed during the rough pump-down, the camera will be in the inserted, open position.

- Put the **CAMERA** switch on the front side of the controller in the **Out** position (Figure 2-2).

  This will keep the camera partially retracted when the viewing chamber pump-down cycle is complete and the diffusion pump gate valve reopens.

- Start the evacuation of the viewing chamber.

Follow the instructions given by the manufacturer.

- When the pump-down cycle is complete, restore the configuration of the pressure lines to the normal configuration as shown in figure 2-5.

Plug tube 1 into **Air Out** on the backside of the controller and tube 3 into tube 6. The camera will remain in the partially withdrawn position until the **CAMERA** switch (Figure 2-3) is set to **Computer** and **INSERT CAMERA** is selected from the Camera menu in Digital Micrograph. Thereafter, the camera will automatically be kept at high vacuum.

A3.3 Water lines

Figure 2-6 shows how the camera is connected to the microscope's water-cooling system. The line used to expel used cooling water from the camera is called the water-exhaust line and the line used to intake cooling water into the camera is called the water-intake line. Lines 1, 2, and 3 are in series and take cooling water to the camera. Lines 4 and 5 return the water to the microscope water chiller.

- Cut the water-chiller hose that runs from the water chiller to the microscope.

The cut must be made before any pressure regulation or flow restriction in the microscope plumbing. This will insure that once the water-intake line is installed, the cooling water will be pushed into the water-intake line so that the water can cool the camera.

- Insert a supplied T into the previously cut hose.

Choose the size of T that best fits the water-chiller hose. Screw-type hose clamps can be used to secure the T to the two ends of the hose.

- Cut the water-chiller hose that runs from the microscope to the water chiller.

The cut must be made after any pressure regulation or flow restriction in the microscope plumbing. This will insure that once the water exhaust line is installed, the thermally degraded cooling water will be pushed out of the water-exhaust line so that the water will not return to the camera.

- Insert a supplied T into the water-chiller hose that runs from the microscope to the water chiller.

Choose the size of T that best fits the water-chiller hose. Screw-type hose clamps can be used to secure the T to the two ends of the hose.

- Connect line 5 to the inserted T.

A screw-type hose clamp can be used to secure the line to the hose.

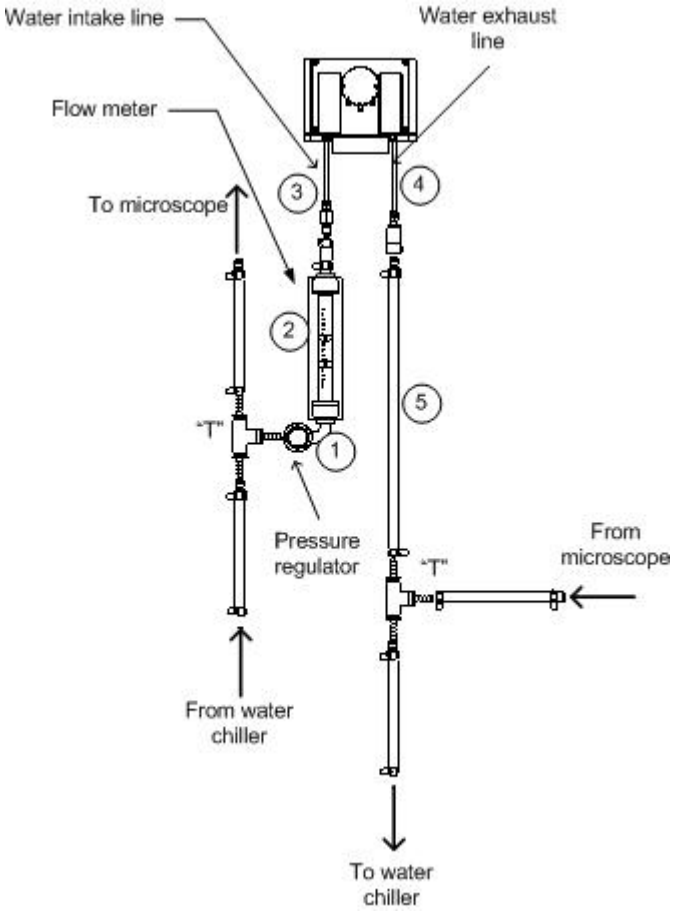
- Set the pressure regulator to obtain a flow of 10-12 L/hr within the limits imposed by the microscope water chiller and the flow needed by the diffusion pump.
- Set the chiller temperature regulator around 20-23 °C.

Note that temperatures lower than 20 °C may result in condensation accumulating in the CCD camera head electronics. This may interfere with normal functioning of the camera.

- Connect line 1 to the pressure regulator end of line 2.
- Connect line 2 (flow meter end) to line 3 and the other end of line 3 to the water inlet on the camera head.
- Connect one end of line 4 to the water outlet on the camera head and the other end to line 5 to complete the loop.

The quick disconnect fittings automatically seal the lines off when not connected to anything, so that they may be connected and disconnected from the camera without affecting the flow of cooling water to the diffusion pump.

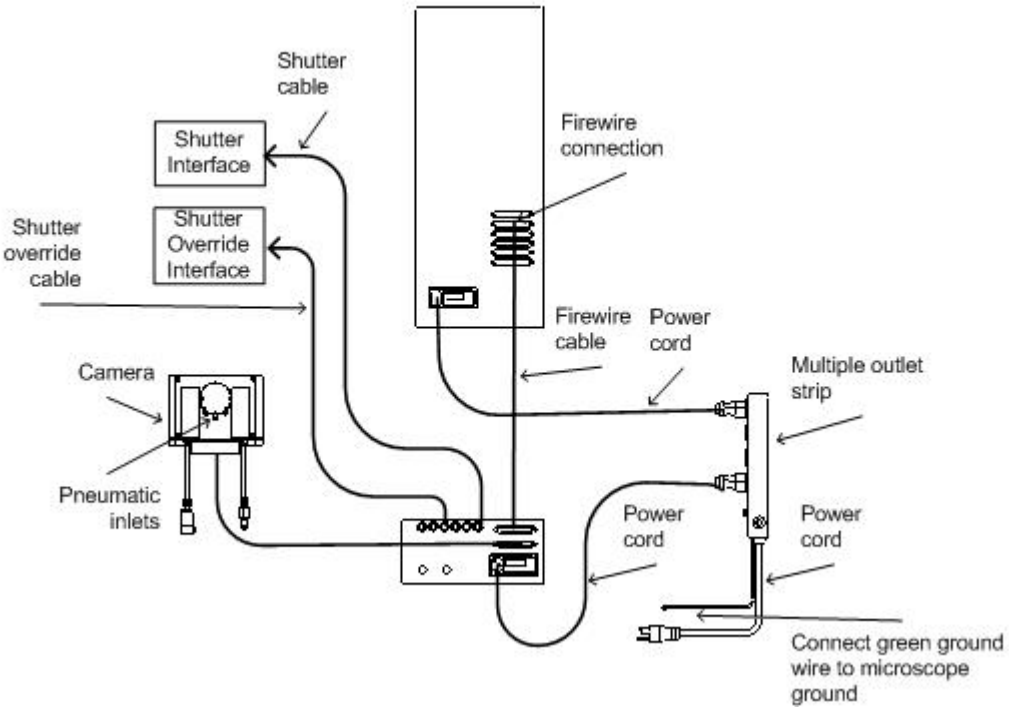
Figure A6 **Water lines**



A3.4 Electrical connections

The diagram in Figure 2-7 shows the electrical connections in the system.

Figure A7 **Electrical diagram.**



- Connect the controller and the computer to the multiple outlet strip.
- Run the camera cable between the camera and the controller.

Always make sure that the controller is switched off when installing or removing this cable.

- Connect the green ground wire on the outlet strip to the microscope ground.
- Run the Firewire cable between the controller and the computer.

A3.5 Shutter-control output

The Shutter-Control output (shown in figure 2-7) is on a BNC connector on the back panel of the controller (Figure 2-3).

- Connect one end of the Shutter-Interface cable to the Shutter-Control output on the controller.
- Connect the other end to the Shutter-Interface provided by Gatan (or microscope vendor).

A3.6 Shutter-override output

The shutter-override output (shown in figure 2-7) is on a BNC connector on the back panel of the controller (Figure 2-3).

- Connect one end of the shutter override interface cable to the shutter override output on the controller.
- Connect the other end to the shutter-override interface provided by Gatan (or microscope vendor).

Appendix 2

CE certification

DECLARATION OF CONFORMITY

(According to ISO/IEC GUIDE 22 and EN 45014)

Manufacturer's name: ***Gatan Inc.***

Manufacturer's Address: ***5933 Coronado Lane***
Pleasanton, CA 94588 U.S.A

DECLARES THAT THE PRODUCT

Products name: ***UltraScan 4000 SP***

Model Number: ***890***

CONFORMS TO THE FOLLOWING EUROPEAN DIRECTIVES

Low voltage Directive 73/23/EEC
EMC Directive 89/336/EEC
As modified by Directive 93/68/EEC

Supplementary Information:

Safety: **EN 61010-1:1991/A2:1995**

EMC: ***EN 55022: Class A/Class B***
EN 55082-1:1992
EN 55081-1:1992

I, the undersigned, hereby declare that the equipment specified above conforms to the above Directives and Standards.

Place: Pleasanton, CA

Date: September 1999

Signature: R. Buchanan

Full Name: Robert Buchanan

Position: President and CEO

Index

A

arrows, **3-3**
Auto Exposure, **3-2**, **3-4**, 4-1
Auto Survey, **3-2**, 4-1

B

binning, 5-3, 5-4

C

Calibrate image, 2-7, **2-9**
calibration, 2-5, 2-7, 2-8, **2-10**
Camera menu, 2-3, 2-4, 4-1, **5-5**
CCD, i, 1-1, 2-1, 2-3, 2-4, 2-5, **2-8**, 3-1, **3-2**, **3-4**, 3-9, 4-1, 5-1, 5-2, 5-3, 5-4, 5-5, I, II
CE certification, XIII, XV
Configure Camera, **2-3**
Custom item, **3-7**, **3-8**, 3-9
customization, 2-1

D

defect correction, 2-3
diffraction patterns, 5-2
digital camera, 1-1, 2-1
DigitalMicrograph, i, ii, 1-1, 2-1, 2-2, 2-5, 3-9, 4-1
dynamic range, 1-1, 5-3

E

exposure time, **3-2**, 5-2, 5-4

F

FFT, 2-5
fiber-optic plate, II
film, 1-1, 2-5, 5-2, 5-4, **5-5**, II
Focus Loupe, **3-2**, **3-3**, 4-1
Focus mode, 3-1

G

gain reference, 2-4, 4-1, 5-4, **5-5**
ghost image, **5-5**
Global Info, **3-4**, 4-1
Global Microscope Info, 2-5, 2-6

H

hardware, I

I

Image Acquisition, i, 2-1, **3-1**, **3-4**
Image Info, **3-3**, **3-4**, 4-1
Image rotation, 2-3
insert/retract, **5-5**

L

linearity, 1-1

M

magnification, 2-5, 2-6, 2-7, 2-8, **2-10**
Magnification, i, 2-5, 2-6, **2-8**
magnification table, **2-8**
Microscope menu, 2-5, 2-6, **3-4**

N

Name field, 2-3
noise, 1-1, II

P

Peltier, 2-1, 4-1, 5-1, 5-2, 5-3
print, 3-8, 3-9, 4-2

R

Reference, i, 1-1, 2-4, 4-1
region of interest, 3-1
Report Options, **3-6**, 3-8
resolution, 1-1, **2-9**, 3-1, **3-4**, 5-3, II
retraction, 1-1, 5-4, II

S

saturation, 5-2, 5-3
Saving, 2-1, **3-3**, **3-5**, **3-6**, **3-7**, 3-9, 4-2
scintillator, 5-2, 5-3, **5-5**, II
Search mode, 3-1, **3-2**, 4-1
sensitivity, 1-1
Set up, **3-5**
shutter, 2-3, 5-3, 5-4, 5-5
speed of operation, 1-1
Standard, 1, 1-1, I, II, III
Start Acquire, **3-4**, 4-1
Start View, **3-2**, 4-1

T

TEM, 1-1, 2-5, **2-8**, **3-4**, **3-7**, **3-8**, 4-1, 5-1, 5-2, 5-4, **5-5**, II, III
transmission electron microscope, 1-1

V

vacuum, 2-1, 4-1, 5-2, 5-3, 5-5
view mode, 3-1